

GAIN OF FUNCTION MUTATIONS IN
ATP-DEPENDENT TRANSPOSITION PROTEINS

ABSTRACT OF THE DISCLOSURE

5 The invention is specifically directed to efficient, random, simple insertion of a
transposon or derivative transposable element into DNA *in vivo* or *in vitro*. The invention
is particularly directed to mutations in ATP-utilizing regulatory transposition proteins that
permit insertion with less target-site specificity than wild-type. The invention encompasses
gain-of-function mutations in TnsC, an ATP-utilizing regulatory transposition protein that
10 activates the bacterial transposon Tn $\underline{7}$. Such mutations enable the insertion of a Tn $\underline{7}$
transposon or derivative transposable element in a non-specific manner into a given DNA
segment. Insertion can be effected in plasmid and cosmid libraries, cDNA libraries, PCR
products, bacterial artificial chromosomes, yeast artificial chromosomes, mammalian
artificial chromosomes, genomic DNAs, and the like. Such insertion is useful in DNA
15 sequencing methods, for genetic analysis by insertional mutagenesis, and alteration of gene
expression by insertion of a given genetic sequence.

20